COMPARISON OF THE FLEISCHL, THE GOWERS AND THE SPECIFIC GRAVITY METHODS OF DETERMINING THE PERCENTAGE OF HÆMOGLOBIN IN THE BLOOD FOR CLINICAL PURPOSES.

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Each year the importance of the clinical examination of the blood is becoming better recognized. In this examination there are two points to be ascertained which are generally acknowledged. These are, the percentage of Hæmoglobin and the number and kind of red and white blood corpuscles.

For determining the hæmoglobin there are several methods. The hæmometer of Fleischl, the hæmoglobinometer of Gowers and the spectroscopic method of Henocque, are fairly well known. None of the above methods employ the microscope, but a determination of the hæmoglobin is so intimately connected with a microscopical examination of the corpuscles of the blood, that we feel justified in presenting this paper.

It is recognized that there is a relation between the specific gravity of the blood and its percentage of hæmoglobin. Hammerschlag has constructed a table giving the hæmoglobin percentages corresponding to the different specific gravities of the blood.

Under the direction of Dr. Williams, professor of pathology in the University of Buffalo, we have made observations upon over 100 patients in the Buffalo General, the Erie County and the State hospitals.

In these observations we have compared the specific gravity method of Hammerschlag with the hæmoglobinometer of Gowers and the hæmometer of Fleischl.

Fleischl's hæmometer consists of a colored wedge, with a graduated scale attached; a well with two compartments, one for

pure water and the other for diluted blood; and a capillary pipette for measuring the blood. The blood obtained, by puncturing the finger, is drawn by capillarity into the pipette, from which it is washed into one of the chambers of the well.

Here it is thoroughly mixed with the water. Both compartments are then filled with water and the well is covered by a glass plate. The well is placed upon the stand so that the compartment filled with distilled water is over the colored wedge. This is moved by a screw until its color corresponds to that of the diluted blood in the other compartment. The percentage of hæmoglobin is then read off from the attached scale. In using the Fleischl, artificial light is necessary, daylight being excluded.

The hæmoglobinometer of Gowers is usually manufactured with but one colored tube, which is for use with daylight. There is another form in which there are two tubes, one for use with daylight and the other for artificial light. The one which we have used is of the former kind. It consists of a sealed tube filled with a glycerine-jelly solution of carmine and picro-carmine of the color of an one-per-cent. solution of normal blood; another tube of the same diameter to hold the blood to be tested; a pipette graduated to 20 cu. m.m. and a stand to hold the two tubes, side by side. The blood measured in the pipette is mixed with a small quantity of water in the graduated tube; water is then added until the dilution corresponds in color to that of the standard solution in the other tube. In making the comparison it is necessary to hold the instrument against a white back ground, opposite the source of light or directly between the eye and the window.

The method which we have used for determining the specific gravity, and thus the hæmoglobin of the blood, is not so well known as the above and will therefore bear a more detailed description. It is one used by Hammerschlag and depends upon the well-known physical principle that a body which will float indifferently in a liquid is of the same specific gravity as that liquid. For this purpose, two liquids are taken, one of a higher and the other of a lower specific gravity than that of the blood, with neither of which it will mix. The necessary apparatus con-

sists of a hydrometer, hydrometer jar, chloroform and benzole.

In using this method, the finger is pricked and the blood thus obtained is introduced into a mixture of chloroform and benzole in the hydrometer jar. The drop of blood, since it will not mix with either chloroform or benzole, retains its spherical form. If the drop sinks the mixture is too light and must be made heavier by adding chloroform. If it rises the mixture is too heavy and must be made lighter by adding benzole. By carefully adding one or the other a point is reached where the drop of blood will neither rise nor sink, but will float indifferently in the mixture. At this point the specific gravity of the blood is the same as that of the mixture. By means of the hydrometer we can obtain the specific gravity of the mixture and thus at the same time that of the blood.

It is desirable to use a medium-sized drop of blood and it is better not to divide this into several. Care must be taken, however, to mix the liquids thoroughly by stirring with the glass rod. In order to facilitate mixing, it is well, when the liquid is too heavy, to add an excess of benzole and bring it back to the desired point by adding chloroform. The latter being heavier, sinks and thus mixes more readily with the mixture.

We have found it convenient to obtain the blood from the palmar surface of the middle finger of the left hand, and have used, for this purpose, an ordinary sharp-pointed steel pen with one nib broken off. A new pen may be used for every test and should be sterilized by heat. The finger also should be washed with some antiseptic, in order to take every precaution against infection. This method of obtaining the blood was used by us for the three instruments.

For introducing the blood into the chloroform-benzole mixture, a pipette of fine calibre may be used. A sufficient quantity of blood is drawn into this and expelled in the middle of the mixture. Care should be taken that all of the blood is not blown out, but that some remains in the tip of the pipette. That which has been expelled will usually adhere to the pipette as a large drop and must be shaken loose. By thus holding back a small portion of

blood, the liability of mixing air with the drop is avoided as much as possible.

E. Lloyd Jones, of Cambridge University, uses a modification of the method of Professor Roy. This, which depends upon the same principle as the preceding, consists in the use of numerous solutions of glycerine and water, the specific gravities of which are known and which are successfully tried until one is obtained, corresponding in specific gravity to that of the blood.

His apparatus consists of twenty to twenty-five one-ounce glass bottles filled with standard solutions of glycerine and water, differing one from the other by .001 of specific gravity; a number of fine glass pipettes drawn out to a point and bent at right angles near the tip; a cylindrical glass jar of about one dram capacity; and a number of clean, sharp suture needles. After puncturing the finger on the dorsal aspect near the root of the nail, the blood which exudes of itself or after the finger has been quickly squeezed, is drawn into one of the pipettes. This is introduced into one of the standard solutions and the blood gently blown out. The solution chosen is of high or low specific gravity according to the appearance of the patient. The bent point of the pipette prevents the blood from being given an impetus up or down when blown from the end.

According to whether the specific gravity of the blood is equal to, greater, or less than that of the solution, it will pursue a horizontal course, sink or rise. By trying a number of solutions one may be found in which the blood neither rises nor sinks, or two are found in one of which it rises and in the other sinks. In the last case the specific gravity of the blood is between the two.

In our experience with the Gowers' instrument, we have found it very unsatisfactory. It is often quite impossible to get the tint of the diluted blood to correspond to that of the standard one-per-cent solution. Even when this is attained, a difference in shade may be produced by looking at the instrument somewhat from the side instead of straight from in front; by holding the paper for reflection farther away from or nearer to the instrument; by holding the instrument between the eye and the window or by

moving farther away from the window. In the last case, in several instances, the differences produced by moving twenty feet away from the source of light, was fifteen per cent., the blood requiring to be more diluted when farther from the window and thus giving a higher reading. These tests were made in a hospital ward on a day of average brightness. Therefore it may be seen that in addition to the other sources of error, the nature of the day, whether it be bright or cloudy, will make an appreciable difference.

We have frequently disagreed in our readings of the same test in both Fleischl and Gowers and others also have differed from us as to when the proper shade was attained. In using the Fleischl instruments, in comparison in the same cases, we have generally found a difference in reading between the two. In thirty per cent. of these comparisons the difference was as much as ten per cent. We have also found that in one-fifth of our cases we disagree in our readings of the same instrument.

We have found it a great inconvenience in making bed-side tests in a hospital ward, to run to some other part of the ward or building to a dark room. In order to obviate this difficulty we have adopted the following device: This consists in our instrument bag fitted with a cardboard cover; at one end of this a hole is cut for the passage of a lamp chimney; at the other end a small hole for looking through the well of the instrument, and at one side of this a window with a flap for inserting the hand to move the wedge.

Hammerschlag's method has the advantage that there is no color test. Every one must agree as to whether the drop rises or sinks or stays where placed. It is also very inexpensive, all that is necessary being a hydrometer jar, chloroform and benzole. The method of Roy and Jones necessitates keeping on hand a large number of solutions which require careful standardization and must be re-standardized at frequent intervals. Although this method may be better where a large number of cases are to be examined in a short time, yet for the ordinary observer, who uses a method of this kind less often and upon a small

number of cases, the one which we have used seems preferable.

In both methods, Hammerschlag and Jones have found that there is no appreciable difference due to variations of temperature in the room.

The results which we have obtained in making parallel tests with the above described methods, may be summarized as follows:

The readings of the Fleischl ran as a rule from ten to fifteen per cent. lower than the percentage estimated from the specific gravity. The readings of the Gowers ran a few per cent. lower than the specific gravity. The Gowers' instrument is liable to an error of at least fifteen per cent. depending upon the intensity of the light. The Fleischl instrument is liable to an error of about ten per cent. In the specific-gravity method there is liability of error from two sources. The drop of blood may adhere to the sides of the jar, or some air may become mixed with it. These errors in the specific-gravity method are reduced to a minimum by careful manipulation.

The greatest error in this last method may be due to the table, since of the cases from which Hammerschlag constructed his table, a great number were primary anæmias and chloroses. For these his table would probably be more accurate than for our cases, as all the anæmias which we examined, were secondary. Our cases were taken as ordinarily found in hospital wards, both medical and surgical, and covered a wide range of diseases.

We are convinced from the experience of others and from our own observations that all of these methods are liable to considerable error. Osler says that the error in the Fleischl instrument may not be more than two per cent. in blood, which is nearly normal, but cites Neubert and Letzius as having shown that in a much impoverished blood the error may be as much as twenty per cent.

The specific-gravity method has the advantage of cheapness and convenience; of taking but little blood, and of not being a color test. This last is of the most importance since the accuracy of the test does not depend so much upon the judgment of the

individual, and makes it practical for observers who lack sufficient appreciation of colors and shades.

In following up a case with a color test, an error of five per cent. too low might be made at the first reading, and one of five per cent. too high at the second and the patient be supposed to have improved to that extent, when, in reality, his condition had remained unaltered. With the specific-gravity method this error is less likely to occur.

It has been found that while the specific gravity may vary at different times of the day, being influenced by sleep, food, drink, exercise, etc., the hæmoglobin, under similar conditions, varies also.

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